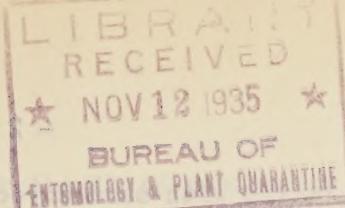


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LABORATORY METHODS OF REARING FOUR SPECIES OF LEPIDOPTEROUS PESTS OF TRUCK CROPS

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The success of toxicological research in entomology is largely dependent on artificial methods of rearing suitable insects for testing purposes. This phase of the work must be thoroughly mastered before any serious work can be undertaken, and the problem often becomes complicated when the work is to be done through the winter in localities where the insects normally hibernate. A reliable method of rearing a suitable species in the laboratory or greenhouse is, therefore, of prime importance to those interested in studies of physiology and toxicology.

The writer has had some experience in rearing insects in the laboratory, attended by reasonable success in the case of the following four species of Lepidoptera: The imported cabbage worm (Ascia rapae L.), the diamond-back moth (Plutella maculipennis Curt.), the southern armyworm (Prodenia eridania Cram.), and the greenhouse leaf tier (Phlyctaenia rubigalis Guen.). The imported cabbage worm was reared throughout the year in the vicinity of Washington, D. C., where it normally hibernates in the pupal stage. The greenhouse leaf tier was also reared for one summer in a laboratory at Washington, but no effort was made to carry it through the winter. All four species were reared successfully in a laboratory at Sanford, Fla.^{1/} A description of the methods used will be given for what they may be worth to others engaged in similar lines of work.

Imported Cabbage Worm

A greenhouse is necessary for rearing the imported cabbage worm because the adults require sunlight for normal activity. The larvae will develop normally without sunlight, but the adults will rarely mate or oviposit in its absence. Incandescent lights have been tried to induce oviposition but with no positive results. An occasional fluctuation in temperature between 50° and 95° can be tolerated by the stock, but it is best to maintain the temperature between 70° and 80° F. The insect is normally a hot-weather pest and develops very nicely at 85° but this temperature is somewhat too high for cabbage plants.

Figure 1 is a drawing based on the system used in rearing the cabbage worm in a greenhouse. The space used is an ordinary plant bed 3 feet wide and from 25 to 30 feet long. The bed is filled with good loam soil, excepting that

^{1/} The writer acknowledges the helpful suggestions of the members of the Sanford, Fla., laboratory of the Division of Truck Crop and Garden Insect Investigations, who were first to carry on laboratory rearing of the southern armyworm and the greenhouse leaf tier.

portion which is filled with sand directly under the cage reserved for larvae. On one end of the bench is an oviposition cage for butterflies which is roughly 2 by 3 by 4 feet in dimensions and has a small door in one end. At each end of this cage a lantana plant is permanently set in the soil to provide flowers on which the adults feed. During about 10 days out of every 60 the plant bears no blossoms, but with two or three plants in the cage blossoms are practically always assured. However, when the plants cease blooming they should be trimmed back to restrict their growth to a reasonable size. Between two lantana plants is an open space reserved for a young potted cabbage plant on which the butterflies lay their eggs. Since the young larva eats the egg case from which it has just hatched, it is very often inclined to continue its feeding on other eggs that have not yet hatched. To hold the destruction of eggs to a minimum, overcrowding of the eggs must be guarded against by changing the plant at least every 24 hours. When the plants are removed from this cage, they are temporarily set in the open bed to await hatching of the eggs.

Seeds are planted periodically in the open bed between the cages to give a continuous supply of young cabbage plants about 5 or 6 inches tall. These are transferred to pots as needed and placed in the oviposition cage. Cabbage is probably best for this purpose, although collards can be used also. Care should be taken to keep the plants free from aphids, as the butterflies do not seem to lay so readily on plants so infested, and the larvae hatching seem to suffer abnormal mortality.

After 3 or 4 days in the egg stage the larvae hatch and are allowed to consume practically the entire plant, which is then broken off at the surface of the ground and laid on top of a large cabbage plant in the larval rearing cage. This cage is 3 feet square and 8 feet long and is covered with wire screen, which should be at least 18 or 20 mesh to exclude spiders and parasites. Two doors in the front of the cage give easy access to any part of it. Large cabbage plants that have not yet headed are dug up from an outside patch or a cold frame and planted in the sand in this cage. When these plants are skeletonized by the larvae they are replaced, care being taken to remove all the larvae from the discarded plants. When full grown, the larvae migrate from the plants and generally pupate on the wire screen at the top of the cage. It was noted that the majority of the larvae pupated in the shady end next to the headhouse. It might, therefore, be well to shade the cage, but it is not recommended that any further hiding place be constructed. If the cage is unobstructed, the pupae can be easily examined and all diseased and parasitized individuals can be removed and destroyed. From healthy pupae adults emerge directly into the larval rearing cage, where they are collected daily and transferred to the oviposition cage for butterflies at the other end of the bench. A cage of this size will accommodate up to 150 adults, but for larger numbers additional cages should be constructed. Even for this number it may be necessary to place two potted cabbage plants in the cage.

If the greenhouse is maintained at a temperature of 75° to 80° F. a complete generation will occur about once a month. In the present work several dozen adults were collected in the field during May, and by August several thousand larvae were available. The insects were maintained in field cages and insectaries during the summer and were transferred to the greenhouse during the

latter part of August. In December, after the third generation in the greenhouse, the pupae entered a resting stage despite the regulated temperature of 75° to 80° F. They remained dormant for about a month and then emerged as healthy adults. The successive generations from these adults developed at the normal rate, suggesting that a resting stage is necessary at the end of a certain number of generations at high temperatures regardless of the season of the year. For this reason it is recommended that field collections be made once a week at the start in order that larvae may be available at all times and the dormant period be more or less covered over by the mixed generations.

Diamond-back Moth

The diamond-back caterpillar is a very important pest on cabbage and other cole crops and is not at all difficult to rear. The larvae develop nicely in ordinary glass battery jars of the 4-quart size, and these may be kept anywhere about the laboratory without regard to sunlight. For steady development of the stock, they should be kept at a fairly constant temperature of about 75° or 80° F.

Figure 2 is a sketch of a method of rearing used satisfactorily at Sanford, Fla. Young potted cabbage plants are necessary, as for the previous species, and these should be grown with the idea of having a constant supply of young plants on hand at all times. When 5 or 6 inches tall, the potted plant is placed in an oviposition cage containing the adult moths. This cage is a cubic foot in its dimensions and is covered with 16-mesh wire screen excepting for the top, which is covered with cheesecloth to diffuse the light. A door sufficiently large to admit a potted plant should be made in the side. The cage is placed in an open greenhouse with the sunlight falling directly upon it, but whether this is necessary was never definitely determined. The adults are not active during the brighter hours of the day and it may be that sunlight is superfluous for their normal activity. Whether there is any necessity for flowering plants as food for the adults was never determined, as no adult moths were ever observed to feed on those placed in the cage.

The eggs are very small, flat discs and are most often laid on the stem portions of the plant rather than on the leaves, although they are often found on the latter. The plants should be changed daily and those bearing eggs should be placed in battery jars with muslin covers held down by rubber bands. If the larvae are allowed to hatch before the plant is placed in a jar, many of them will be lost by dropping from the plant by a spun thread. Just why the larvae do this is uncertain, but it may be because of unnatural crowding. At any rate, placing the potted plant in a battery jar seems to increase the number of larvae developing from the stock of eggs. The muslin cap is necessary on the battery jar because the small size of the larvae enables them to crawl through fabrics of coarser mesh such as ordinary cheesecloth.

After hatching, the larvae feed on the leaves of the cabbage plant, many of them burrowing into the tissues of the leaves and thus disappearing from sight. When they have skeletonized the potted plant, it is cut at the surface

of the ground and the pot removed, leaving the cut plant lying on the bottom of the jar. Fresh cabbage leaves are then introduced in the jar as needed. When the larvae have become full grown they pupate in fine silken nets attached to the cabbage leaves. The jars should now be gone over daily and all pupae removed with a pair of forceps and placed on a false screen bottom inserted in an ordinary battery jar. This screen separates the pupae from the bottom of the glass jar and consequently from any condensation or other water that may find its way into the jar. As the adults emerge they may be collected in any manner convenient to the operator. This may be accomplished by the use of an inverted funnel which is placed over the top of the battery jar and through which the adult moths pass into a small bottle and thus are taken to the oviposition cage.

At temperatures of 75° to 80° F. a generation will develop in about a month. This species was never reared for more than 6 months at a time and consequently it is not known if successive generations can be raised without the intermission of a resting period. However, if a resting stage occurs it can be taken care of by staggering the generations, as in the case of the cabbage worm. There is no reason to think that this species could not be reared in a greenhouse by the same methods and equipment employed for the cabbage worm.

Southern Armyworm

The southern armyworm can be reared in any laboratory without elaborate equipment or technical assistance. It is very resistant to disease and with ordinary care can be raised in large numbers. The insect does well at ordinary room temperatures, especially between 70° and 80° F. The larvae are voracious feeders and can be reared on the leaves of celery, turnip, cabbage, sweetpotato, and a number of other truck crops.

A laboratory method of rearing the armyworm is shown graphically in figure 3. From a dozen to two dozen moths are placed in an ordinary glass battery jar with a cheesecloth cover. Within the jar is also a potted celery plant or merely cut leaflets placed in a flask of water. A young collard or cabbage plant could probably also be used in place of the celery, although celery offers more chance of concealment for the adults during the daylight hours when they are normally not active. The moths begin their activity at twilight, mating and ovipositing readily within the small jar. The eggs are deposited largely on the potted celery plant but may also be deposited on the cheesecloth cover or on the sides of the glass jar. They are light green in color, deposited in masses, and covered with a thin layer of cotton-like down. When oviposition begins, the moths should be transferred daily to a new jar containing a fresh plant. Since the moths are not active in the daylight, this can be done readily by lifting the moths by a leg or wing with a pair of forceps. By holding the egg-bearing plant over the fresh jar, many of the moths can be gently knocked off to the new plant. The new jar is covered with cheese-cloth and set aside for another day while the old jar is set aside for hatching of the eggs. In a little less than a week at 75° the larvae hatch and very shortly consume the plant. Often up to several thousand larvae will be found in a single jar and in such cases they should be thinned out into other jars. New foliage should be placed in the jars as it is consumed by the larvae. When

the larvae have become half grown they should be changed occasionally to a clean jar. When some of the larvae have entered the last instar they should be changed to a jar containing from $1\frac{1}{2}$ to 2 inches of moist sand. The leaves are placed on the sand, and as the larvae finish feeding they burrow down into the sand to pupate. When all the larvae have pupated the debris is cleaned from the jar, and after a period of 3 or 4 days the sand is dumped from the jar and pupae are recovered. These are laid on the surface of a layer of moist sand in another jar and set aside for emergence.

A complete generation of this insect develops in about a month at 70° to 80° . A resting stage does not seem to be necessary for this species as it was in the case of the cabbage worm. The battery-jar method of handling the larvae is probably not the best one. Some such arrangement as was used for the cabbage worm might give better results and require less labor.

Greenhouse Leaf Tier

The greenhouse leaf tier is much more susceptible to disease than is the armyworm, but with a reasonable amount of care it can be raised in large numbers. A temperature of 70° to 75° F. is probably best for the leaf tier. Generations will occur about once a month at this temperature, and there is no apparent rest period as in the case of the cabbage worm.

In rearing the species the same method was used as for the diamond-back moth. Therefore, figure 2 will also illustrate the necessary procedure for the leaf tier. In this case, however, small potted celery plants were used rather than cabbage, and celery leaflets were fed the larger larvae instead of cabbage or turnip. The larvae, pupae, and adults were handled in exactly the same way for both species.

It is possible that the leaf tier can be reared almost free of disease providing some such method is used as is described for the cabbage worm. The elimination of battery jars and substitution of a wire-screen cage for the larvae is a step in the right direction for any insect that is susceptible to disease. The free circulation of air and lower humidity discourages the spread of the disease. It should be pointed out, however, that the battery-jar method is almost indispensable where work is being done that requires several thousand larvae of known age at one time. What should be done in this case is to place some of the plants bearing eggs in battery jars to be used later for testing purposes and keep the rearing stock in an open cage where healthier conditions prevail. In general, leaf tier larvae are not greatly affected by disease until the last instar, and this difficulty may be overcome by using younger larvae for experimental purposes.

List of Illustrations

Figure 1.--Laboratory method of rearing the imported cabbage worm.

Figure 2.--Laboratory method of rearing the diamond-back moth and the greenhouse leaf tier.

Figure 3.--Laboratory method of rearing the southern armyworm.

When young cabbage is nearly skeletonized it is cut at surface of ground and laid on top of large cabbage plant in rearing cage. Larvae feed on large plants until grown and then pupate on wire screen at top of cage.

On emergence, adults are placed in wire-screen cage to oviposit on young potted cabbage placed between flowering lantana plants.

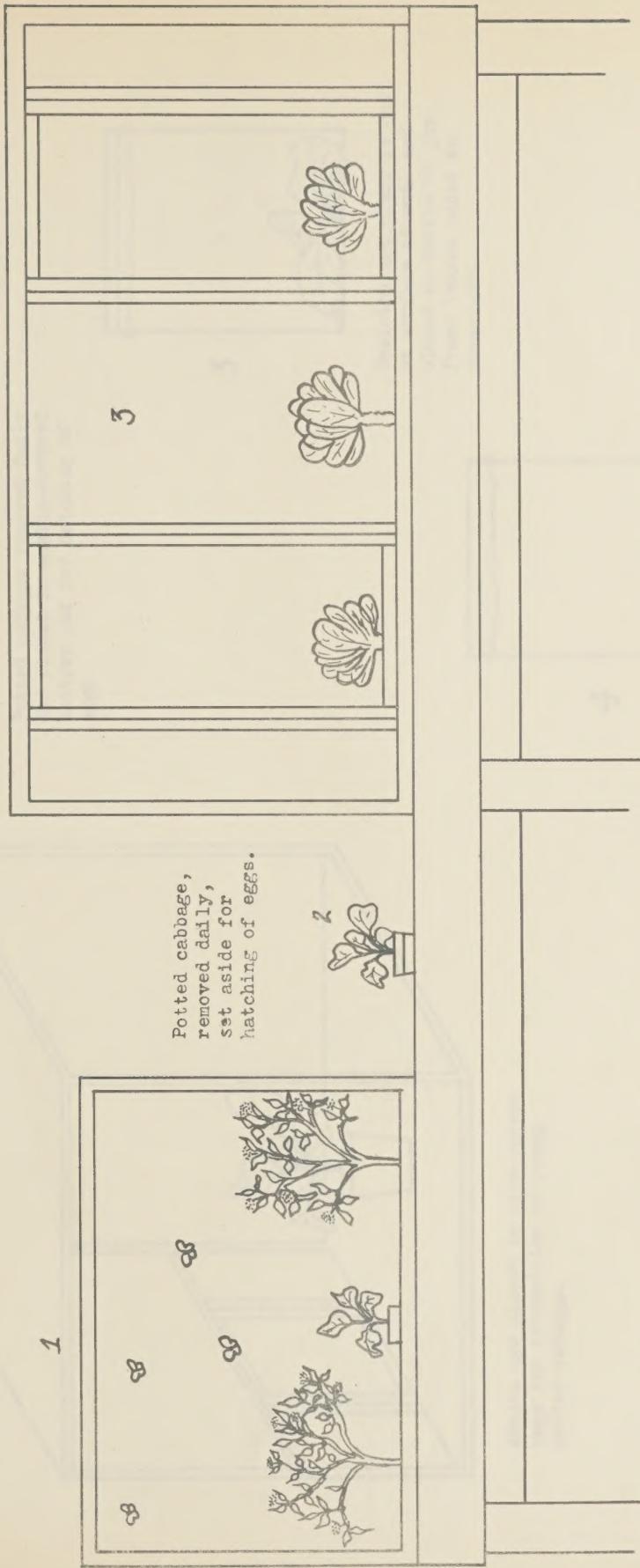
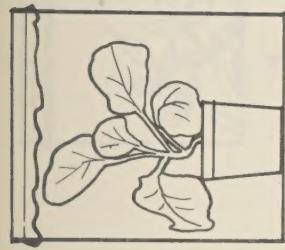
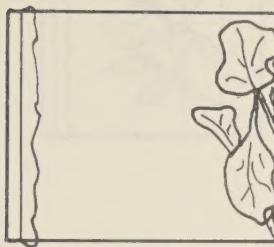


FIGURE 1.



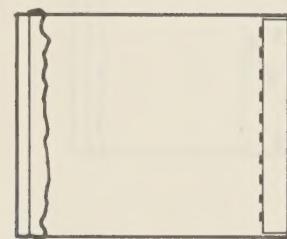
2

Potted cabbage removed daily
and placed in muslin-covered
battery jar for hatching of
eggs.



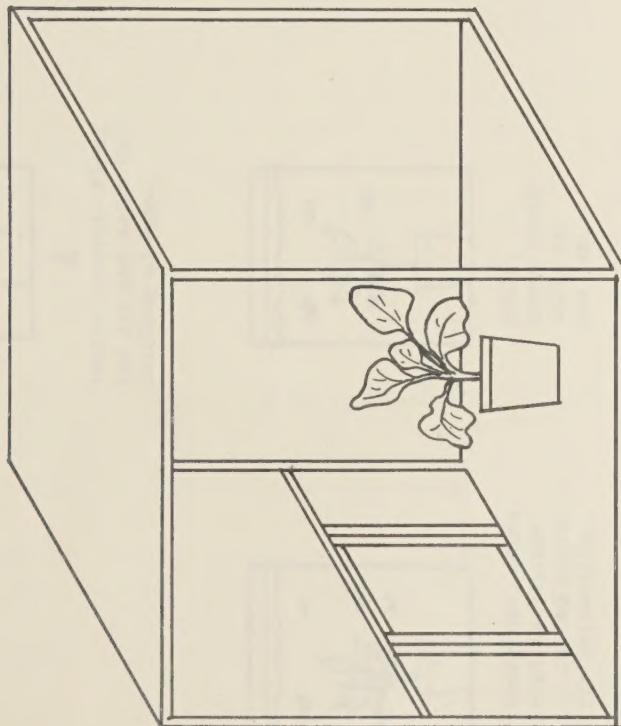
3

Skeletonized plant is cut
at surface of soil and
placed on bottom of jar,
fresh leaves added as
necessary.



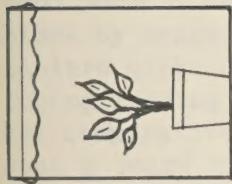
4

Pupae are removed from rearing
jar and placed on wire-screen
support in another jar to await
emergence.

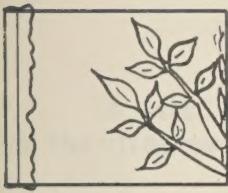


Adults are placed in wire-screen
cage for oviposition on young
potted cabbage.

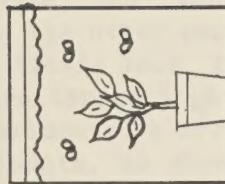
FIGURE 2.

**2**

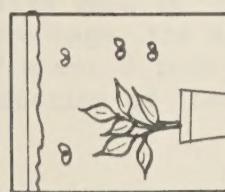
Adults removed- Plant and jar set aside for hatching of eggs.

**3**

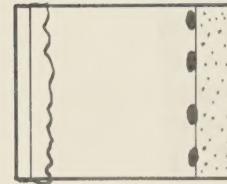
When plant is skeletonized,
pot is removed and fresh
leaves introduced.

**4**

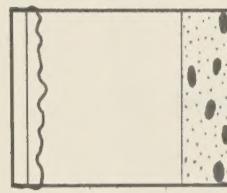
When larvae are about
grown moist sand is
placed in jar.



After emergence, moths
are placed in battery
jar with potted celery
plant for oviposition.

**6**

Pupae removed from sand
after 5 days and placed
on surface of moist sand
in another jar.

**5**

Larvae mature in moist
sand and debris is re-
moved from jar.

FIGURE 3.

